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SHORT COMMUNICATION

Laboratory Demonstration of Oviposition by *Aedes aegypti* (Diptera: Culicidae) in Covered Water Jars

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ABSTRACT In Thailand, ceramic jars (135-200 liters) covered by aluminum lids commonly are used to store water for household use. In laboratory cages, gravid female *Aedes aegypti* (L.) were able to enter and oviposit in a covered water jar. Although the aluminum lid was not a complete barrier to gravid females, it reduced the number of eggs oviposited by 77%. A vertically oriented foam rubber ring which was compressed between the lip of the jar and the flange of the lid effectively sealed the jar against ovipositing mosquitoes.

KEY WORDS *Aedes aegypti*, oviposition, water-jar covers

Aedes aegypti (L.) is a vector of dengue virus in rural areas of Thailand (Sheppard et al. 1969, Halstead 1984, Chareonsook et al. 1990), where roughly glazed ceramic "ong" jars (135-200 liter capacity) are an important larval habitat for this species. We observed an average of six water jars per house in a rural Thai village, and 44% of these jars were infested with *Ae. aegypti* larvae (Kittayapong & Strickman 1993).

Jars often are covered with a commercially manufactured aluminum lid to keep the water clean. These lids have been promoted in Thailand as a means of preventing the larval development of *Ae. aegypti* (e.g., Prugsaphong 1990), because they appear to form a solid barrier against mosquitoes. Contrary to expectation, a recent survey of larval sites recorded a greater proportion of infestation in covered than in uncovered jars located outdoors (Kittayapong & Strickman 1993). Our laboratory experiment measured the ability of gravid *Ae. aegypti* to enter a water jar beneath modified and unmodified commercial covers.

Materials and Methods

The strategy of this experiment was to count the number of eggs oviposited in a covered water jar exposed in a cage to five gravid *Ae. aegypti* during 24 h, then to count the number of eggs

oviposited by the same mosquitoes after a second 24-h period during which the jar was uncovered. Comparison of the number of eggs oviposited on the first and second days provided a measure of the degree to which a cover discouraged oviposition.

The mosquitoes used in this study were the F_1 progeny of larvae collected in a Thai village (Hua Sam Rong Subdistrict, Plaeng Yao District, Chachoengsao Province). Eggs for each cohort were hatched simultaneously under partial vacuum (Barbosa & Peters 1969) and reared under closely controlled environmental (30°C, equal hours of light and dark) and nutritional (50 larvae per 1.5 liters with 1.5 g ground fish food during development) conditions. Five-day old females that took a full human blood meal were incubated for 47-49 h at 30°C with access to a cut raisin before they were released into a test cage with the experimental jar.

The experimental jar was located in a nylon-tulle cage (1 by 1.5 by 2 m high) in a room open to ambient temperature in Bangkok. Water temperatures in the jar ranged between 27 and 31°C during the experiment; 29°C was the median temperature. The experimental water jar was similar to those used in rural Thailand (200-liter capacity, 71 cm high, 43 cm inner diameter at opening), with no holes or large gaps on the lip or body of the jar. The water jar was filled about one-third full with ≈ 80 liters of water.

During each trial, strips of kraft paper towels (8 cm vertical, 25 cm horizontal) were taped to the inner wall of the jar so that the water line covered the lower edges and the sides of the strips overlapped. Depending on the trial treatment, the appropriate cover (or no cover) was placed on the jar, then five gravid mosquitoes were introduced into the cage. After 23.5-24.5 h, the paper towel strips were replaced with new

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Fig. 1. Modifications of commercial aluminum lids with vertically oriented (left) and horizontally oriented (right) rings of foam rubber. The lids rest on a Thai water jar.

strips for the second day of the trial, the jar being left uncovered. After the second day (47.5–48.5 h), strips were again removed and mosquitoes were captured and killed in a small net. Eggs on the strips were counted immediately after they were removed from the jar.

Each of the following four treatments was replicated six times: (1) an unmodified commercial aluminum cover; (2) a commercial cover modified by gluing (Dunlop Adhesive LP, Laminated Plastics Adhesive, Contact Adhesive, Dunlop, Thailand) a horizontally oriented ring of foam rubber (2 cm thick) in the commercial aluminum cover (Fig. 1), forming a layer between the metal lid and the top of the lip of the jar; (3) a commercial cover modified by gluing a 2-cm layer of sponge rubber to the inner side of the flange of a metal cover (Fig. 1), forming a vertical sponge ring between the outer side of the jar lip and the flange of the lid; and (4) the jar left uncovered during both days of each trial.

In addition to noting whether or not eggs were oviposited in the jar, the quantity of eggs oviposited on the first day of each replicate was regressed as a function of the total eggs oviposited during both days and compared among treatments using an analysis of covariance (ANCOVA) (Steel & Torrie 1960). The ANCOVA adjusted for the number of eggs oviposited in each trial. Differences between adjusted treatment means were tested in a series of *t* tests based on the calculation of the standard error of the difference between each pair of means. The uniformity of the total number of eggs oviposited among treatments was analyzed by a one-way ANOVA.

Results and Discussion

Gravid *Ae. aegypti* entered the water jar covered by the unmodified commercial aluminum

Table 1. Effect of various modifications of commercial aluminum covers on oviposition by *Ae. aegypti* in a water jar

Cover treatment	No. trials with ovip. on day 1	Total no. eggs	Mean no. eggs (% ^a) per trial	
			Day 1	Day 2
Unmodified	4	1,135	46 (24)	144 (76)
Horizontal sponge ring	4	935	24 (16)	132 (84)
Vertical sponge ring	0	1,064	0 (0)	177 (100)
No cover	6	1,092	176 (96)	6 (4)

^a Percentage of total eggs oviposited on both days.

lid in four of six trials (Table 1). Evidently, the females were able to find small gaps between the lid and the lip of the jar. Modification of the lid by adding a horizontally oriented layer of foam rubber also failed to prevent entry by ovipositing females in four of six trials. In contrast, no eggs were oviposited when the layer of foam rubber was oriented vertically between the outer side of the jar lip and the inner side of the lid flange.

Differences between treatments were indicated by the numbers of eggs deposited on the first day (adjusted $F = 27.49$; $df = 3, 14$; $P < 0.005$). Significantly fewer ($P < 0.005$) eggs were oviposited on the first day in trials with covers than in trials without a cover. The differences between various covers were not statistically significant ($P > 0.05$). The total number of eggs oviposited during both days of each trial did not vary significantly among treatments (mean, 176 eggs per trial; unadjusted $F = 0.32$; $df = 3, 20$; $P > 0.1$), demonstrating that the differences between treatments were not the result of changes in fecundity induced by the various treatments.

The reduction in number of eggs oviposited when jars were covered can be explained either by each mosquito ovipositing fewer eggs or, more likely, fewer mosquitoes entering the jar. Because only 24 and 16% of the eggs were oviposited when the jar was covered with the unmodified cover and cover with horizontal foam ring, respectively (Table 1), it is probable that only about one in five females entered the jar under these covers. No mosquitoes were able to enter the jar having a cover with a vertical foam ring. Gravid mosquitoes that did not enter the jar and oviposit on the first day with the cover in place evidently held their eggs and then oviposited them on the second day when the cover was removed. Retention of eggs did not reduce the total number of eggs oviposited after the cover was removed.

Modification of metal lids with vertical foam rings may prove to be a practical way to reduce populations of *Ae. aegypti* in Thailand. Gluing the foam into a lid as described in this paper costs only about \$0.15 and takes advantage of the investment in the lid already made by the householder. Use of the lid with a foam ring would be

especially helpful for drinking water in jars that require frequent access and which are not readily treated with chemicals. Field trials that include use of the modified lid are currently in progress in cooperation with the Thai Ministry of Public Health.

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